International Journal of Chemical Studies

P-ISSN: 2349–8528
E-ISSN: 2321–4902
www.chemijournal.com
IJCS 2020; 8(2): 1141-1147
© 2020 IJCS
Received: 06-01-2020
Accepted: 10-02-2020

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Efficacy of fungicides *in vivo* and *in vitro* for the management of head smut caused by *Sorosporium paspali-thunbergii* in Kodo millet

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DOI: https://doi.org/10.22271/chemi.2020.v8.i2r.8922

Abstract
Minimum head smut incidence (0.7%) was recorded in seed treatment with carbendazim@ 2 g kg^{-1} seed (2010-11) whereas maximum grain yield was recorded 1750 kg/ha in seed treatment with carboxin@ 2 g kg^{-1} seed during 2010-11. During 2011-12, minimum head smut incidece 0.7% was recorded in seed treatment with carboxin@ 2 g kg^{-1} seed, where as maximum grain yield was 1710 kg/ha. recorded in seed treatment with carboxin. On the basis of two year pooled data, minimum head smut incidence was 1.0% in seed treatment with carboxin@ 2 g kg^{-1} seed and maximum grain yield 1730.0 kg/ha. was also recorded in seed treatment with carboxin@ 2 g kg^{-1} seed. *In vitro*, average teliospore germination 12.4%, 16.2% and 14.2% was recorded in carboxin, mencozeb and carbendazim, respectively at 100 ppm concentration. At 300 ppm fungicidal concentration, mean teliospore germination 10.1% in carboxin, 12.9% in mencozeb and 11.5% in carbendazim was recorded. Mean teliospore germination 2.8%, 8.3%, 3.1% was recorded in carboxin, mencozeb and carbendazim, respectively at 500 ppm, where as 20.5 to 46.5% teliospore germination was recorded in control. Carboxin was found most effective in reducing the teliospore germination followed by carbendazim and mencozeb.

Keywords: Fungicides, *in vivo*, *in vitro*, *Sorosporium paspali thunbergii*, Kodo millet

Introduction

Kodo millet (*Paspalum scrobiculatum* L.) is one of the hardiest as well as historical crop and indigenous to India (De wet *et al*., 1983) [5] and belonging to family Poaceae (Gamineae). Among small millets generally kodo millet cultivated in low fertile soil by poor peoples for food and feed for animals. The crops is cultivated is about 224 thousands hectares in India with productivity of 312 kg $kgha^{-1}$ (Anon, 2011) [2]. The crop possesses a number of valuable characteristics such as more herbage, branched ear, large number of seeds per ear, high drought tolerance and unique storage ability. Kodo millet has considerable production potential in marginal, low fertility soils and chronic moisture deficient areas of the country. Kodo millet is predominantly grown in Madhya Pradesh, which shares an area of nearly 71.71% to its total area in the country. Uttar Pradesh ranks second followed by Tamil Nadu, Gujarat, Maharashtra, Karnataka and Andhra Pradesh. Kodo millet is nutritionally comparable or even superior to major cereals especially with respect to protective nutrients. Medicinally Kodo plant is styptic used in inflammation, diseases of liver, ulcer, dysentery and heat the body to both human and cattles (Blatter *et al*, 1975) [3].

Besides other limiting factors, head smut caused by *Sorosporium paspali-thunbergii* is an important disease, which is endemic in all the states of the country. Generally all the panicles of an infected plant converted into smut sori and results in corresponding yield reductions. The severity of disease has been reported from 0.0 to 56.4% depending upon the soil, climate and host. The crop is generally grown by tribal and poor farmers in extreme climatic conditions and management of the disease through growing resistant varieties is the cheapest and best way, though few fungicides were found very effective *in Vitro* and *in Vivo*. Some fungicides were also found effective for the management of head smut (Sattar, 1930., Parambaramani *et al*, 1973., Jain and Gupta, 1993., Jain 2004) [18, 17, 10, 8].

Materials and Methods

In Vivo studies

To study the efficacy of commonly available fungicides against head smut of kodo millet, seven fungicides namely Carboxin, Chlarothalonil, Saaf, Mencozeb, Carbendazim, Raxil and Tricyclazole were used as seed dresser to control head smut of kodo millet. Seeds of kodo millet susceptible variety GPUK 3 were inoculated with viable teliospores of the fungus and next day the inoculated seeds were treated with test fungicides @ 2 g kg^{-1} seed. The seeds were sown in a plot size of 2.25 x 3.0 m in three replications during *kharif* 2010-11 and 2011-12. The recommended doses of fertilizers i.e. 40:20:0 kg NPK ha^{-1} were applied for optimum crop growth. Observations on head smut incidence (%) were recorded by counting healthy and smutted plants in each plot at dough stage. Grain yield was recorded at maturity per plot basis and converted into kg per hectare by using the following equation.

$$\text{Grain Yield (kg/ha.)} = \frac{\text{Yield (kg)/ plot x 10000}}{\text{Area of plot}}$$

Treatment details

T_1: Seed treatment with Carboxin @ 2 g per kg seed
T_2: Seed treatment with Chlarothalonil @ 2 g per kg seed
T_3: Seed treatment with Saaf @ 2 g per kg seed
T_4: Seed treatment with Mencozeb @ 2 g per kg seed
T_5: Seed treatment with Carbendazim @ 2 g per kg seed
T_6: Seed treatment with Raxil @ 2 g per kg seed
T_7: Seed treatment with Tricyclazole @ 2 g per kg seed
T_8: Untreated control

In Vitro studies

To study the efficacy of fungicides on teliospore germination of *Sorosporium paspali thunbergii*, three fungicides namely Carbendazim, Carboxin, Mancozeb were used in different concentrations *viz*. 100ppm, 300 ppm and 500ppm.

The suspension of freshly collected viable teliospores was centrifuged at 100 rpm for 2 to 3 minute to get the teliospore pellet. Teliospore pellet were wash with sodium hypochlorite (0.25%) for 45 seconds followed by rinsing twice in sterile distilled water. Pellet was again subjected to centrifugation for two minute at room temperature to remove surfactant and water. Fungicidal solution of desired concentration was prepared and one drop of solution was placed in the cavity slide. Then small quantity of sterilized teliospores was added in the cavity slide containing fungicidal solution. The cavity slides were placed in moist chamber prepared in petri plates and incubated at room temperature for 24 hours. The experiment was conducted in 3 replications. Control was maintained using distil water in place of fungicidal solution. Germination of teliospores was recorded under compound microscope after 3, 4, 5 and 6 days in all the treatments and percent germination was calculated using following Formula:

$$\text{Percent germination} = \frac{\text{Number of spore germinated}}{\text{Total no. of spores observed}} \times 100$$

Percent inhibition in teliospore germination was calculated using following formula:

$$\text{Percent inhibition} = [(X-Y)/X] \times 100$$

Where

X = Spore germination in check
Y = Spore germination in treatment

Results and Discussion

In Vivo studies

Result revealed that the efficacy of evaluated fungicides as seed treatment on incidence of head smut and grain yield of kodo millet during *kharif* 2010-11 was studied and data are presented in table 1 and fig 1. Significant differences among the treatments for percent head smut incidence and grain yield were observed. Head smut incidence varied 0.7 to 11.2% in different treatments. Lowest smut incidence was recorded in seed treatment with carbendazim @ 2 g kg-1 seed (T_5) followed by chlarothalonil (0.8%) and saaf (0.9%). These treatments were observed at par for their effectiveness against head smut. Percent disease control ranging from 7.6 to 15.9% was found maximum in these treatments. Grain yield ranging from 1510.0 to 1750.0 $kgha^{-1}$ was also recorded maximum in the same treatments. An increase in grain yield varied 7.6 to 15.9% in different treatments was maximum in seed treatment with carboxin @ 2 g kg^{-1} seed followed by carbendazim and saaf.

During *kharif* 2011-12, head smut incidence and grain yield varied from 0.7 to 18.7% and 1510 to 1750 kg ha^{-1}, respectively were recorded (Table 2 and Fig. 2). Significant variation among treatments for head smut incidence and grain yield were recorded. Percent smut control and percent increase in grain yield ranging from 70.1 to 96.3% and 7.9 to 17.1%, respectively were recorded in different treatments. Lowest head smut incidence with maximum disease control and highest grain yield was recorded in seed treatment with carboxin @ 2 g kg^{-1} seed followed by carbendazim and saaf. Least smut control was recorded in seed treatment with mencozeb (5.6%), followed by tricyclazole (5.5%) with 9.3% and 7.6% increase in grain yield in comparison to control.

The results on efficacy of fungicides against head smut and grain yield of kodo millet on the basis of two year pooled data are presented in table 3 and fig. 3. Average head smut incidence and percent smut control ranging from 1.0 to 14.9% and 59.7 to 93.3%, respectively were recorded in different treatments. Minimum head smut incidence with maximum disease control was recorded in seed treatment with carboxin @ 2 g kg^{-1} seed followed by carbendazim (1.2% and 91.9%), saaf (1.5% and 88.9%) and chlarothalonil (1.7% and 88.6%). Least head smut incidence and percent disease control was recorded in seed treatment with mencozeb @ 2 g kg^{-1} seed (7.0% and 59.7%) followed by tricyclazole (5.9% and 60.4%). Grain yield varied from 1485.0 to 1730 $kgha^{-1}$ and percent increase over control from 7.7 to 16.5% was maximum in seed treatment with carboxin @ 2 g kg^{-1} seed followed by carbendazim (1717.5 $kgha^{-1}$ and 15.7%), Chlarothalonil (1687.5 $kgha^{-1}$ and 13.6%) and saaf (1675.5 kgha-1 and 12.8%).

Minimum head smut incidence (0.7%) was recorded in seed treatment with carbendazim@ 2 g kg^{-1} seed during 2010-11. Maximum grain yield was recorded 1750 kg/ha in seed treatment with carboxin@ 2 g kg^{-1} seed during 2010-11. During 2011-12, minimum head smut incidece 0.7% was recorded in seed treatment with carboxin@ 2 g kg^{-1} seed, where as maximum grain yield was 1710 kg/ha. recorded in seed treatment with carboxin. On the basis of pooled data, minimum head smut was 1.0% in seed treatment with carboxin@ 2 g kg^{-1} seed and maximum grain yield 1730.0 kg/ha. was also recorded in seed treatment with carboxin@ 2 g kg^{-1} seed. Parambaramani *et al*. (1973) [17] also reported similar findings. Smut in barnyard millet (Pall and Nema, 1978) [16], smut in foxtail millet (Kulkarni *et al.* 1979) [13], head smut in kodo millet (Pall, 1985; Chalam *et al.* 1989: Jain and Gupta, 1993; Jain 1995; Mantur *et al.* 1997; Jain 1999; Jain 2004a) [15, 4, 11, 6, 14, 7, 9], smut in proso millet (Sharma and Sugha, 1991), *Sporisorium destruens* in millet (Kovacs *et al.* 1997) [12] were also reported the efficacy of fungicides in the management of smuts.

In Vitro*, efficacy of fungicides on teliospore germination of *Sorossporium paspali thunbergii

Observations of teliospore germination along with% inhibition as influenced by three fungicides namely carboxin, mencozeb and carbendazim at 3 concentrations i.e. 100, 300 and 500 ppm were recorded after 3, 4, 5 and 6 days and data are presented in table 4, fig. 4, 5 and plate 1. Significant variation in spore germination among different treatments were recorded. Mean teliospore germination ranging from 7.5 to 18.7%, 11.8 to 22.4% and 9.8 to 20.3% was recorded in carboxin, mencozeb and carbendazim, respectively at 100 ppm concentration. At 300 ppm fungicidal concentration, mean teliospore germination ranging from 5.6 to 14.5% in carboxin, 8.9 to 18.8% in mencozeb and 7.5 to 16.8% in carbendazim was recorded. Teliospore germination ranging from 0.0 to 9.0%, 3.3 to 14.8%, 0.0 to 9.6% was recorded in carboxin, mencozeb and carbendazim, respectively at 500 ppm, where as 20.5 to 46.5% teliospore germination was recorded in control. Among the fungicides, carboxin was found most effective in reducing the teliospore germination followed by carbendazim and mencozeb. Least spore germination was recorded at 500 ppm in all the tested fungicides i.e carboxin (2.8%), carbendazim (3.1%) and mencozeb (8.3%). Percent inhibition of teliospore germination ranging from 49.8 to 91.3% was recorded in different treatments over control. Maximum inhibition was recorded in carboxin (91.3%), followed by carbendazim (90.4%) and mencozeb (74.3%) at 500 ppm.

Mean teliospore germination 12.4%, 16.2% and 14.2% were recorded in carboxin, mencozeb and carbendazim, respectively at 100 ppm concentration. At 300 ppm fungicidal concentration, mean teliospore germination 10.1% in carboxin, 12.9% in mencozeb and 11.5% in carbendazim was recorded. Mean teliospore germination 2.8%, 8.3%, 3.1% was recorded in carboxin, mencozeb and carbendazim, respectively at 500 ppm, whereas 20.5 to 46.5% teliospore germination was recorded in control. Carboxin was found most effective in reducing the teliospore germination followed by carbendazim and mencozeb. *In vitro*, carboxin was effective against *Sorosporium paspali-thunbergii* also observed by Ahmad *et al.* (1994).

Table 1: Efficacy of fungicides on incidence of head smut and grain yield in kodo millet during *kharif* 2010-11

Code	Treatments	Head smut (%)	% disease control	Grain Yield (Kg/ha)	% increase over control
T_1	Seed treatment with Carboxin @ 2 g per kg seed	1.2(6.28)*	89.3	1750.0	15.9
T_2	Seed treatment with Chlarothalonil @ 2 g per kg seed	0.8(5.20)	92.9	1695.0	12.3
T_3	Seed treatment with Saaf @ 2 g per kg seed	0.9(5.52)	92.0	1690.0	11.9
T_4	Seed treatment with Mencozeb @ 2 g per kg seed	6.3(14.50)	43.8	1650.0	9.3
T_5	Seed treatment with Carbendazim @ 2 g per kg seed	0.7(4.89)	93.8	1745.0	15.6
T_6	Seed treatment with Raxil @ 2 g per kg seed	1.9(7.92)	83.0	1680.0	11.3
T_7	Seed treatment with Tricyclazole @ 2 g per kg seed	6.2(14.46)	44.6	1625.0	7.6
T_8	Untreated control	11.2(19.54)		1510.0	
	Mean	3.7(11.09)		1668.1	
	S.Em±	0.4		13.54	
	CD (5%)	1.23		41.01	
	CV (%)	7.18		1.40	

* Figures in parentheses are arc sin transformed values

Table 2: Efficacy of fungicides on incidence of head smut and grain yield in Kodo millet during *kharif* 2011-12

Code	Treatments	Head smut (%)	% disease control	Grain Yield (Kg/ha)	% increase over control
T_1	Seed treatment with Carboxin @ 2 g per kg seed	0.7(4.80)*	96.3	1710.0	17.1
T_2	Seed treatment with Chlarothalonil @ 2 g per kg seed	2.6(9.28)	86.1	1680.0	15.1
T_3	Seed treatment with Saaf @ 2 g per kg seed	2.0(8.13)	89.3	1661.0	13.8
T_4	Seed treatment with Mencozeb @ 2 g per kg seed	5.6(13.69)	70.1	1580.0	8.2
T_5	Seed treatment with Carbendazim @ 2 g per kg seed	1.7(7.49)	90.9	1690.0	15.8
T_6	Seed treatment with Raxil @ 2 g per kg seed	2.8(9.63)	85.3	1650.0	13.0
T_7	Seed treatment with Tricyclazole @ 2 g per kg seed	5.5(13.56)	70.6	1575.0	7.9
T_8	Untreated control	18.7(25.62)		1460.0	
	Mean	4.9(12.79)		1625.8	
	SEm±	0.626		23.02	
	CD (5%)	1.89		69.41	
	CV (%)	9.39		2.45	

* Figures in parentheses are arc sin transformed values

Table 3: Efficacy of fungicides on incidence of head smut and grain yield in kodo millet pooled over year, 2010-11 and 2011-12

Code	Treatments	Head smut (%)		Mean	% disease control	Grain Yield (Kg/ha)		Mean	% increase over control
		2010-11	2011-12			2010-11	2011-12		
T_1	Seed treatment with Carboxin @ 2 g per kg seed	1.2(6.28)*	0.7(4.80)	1.0(5.74)	93.3	1750.0	1710.0	1730.0	16.5
T_2	Seed treatment with Chlarothalonil @ 2 g per kg seed	0.8(5.20)	2.6(9.28)	1.7(7.49)	88.6	1695.0	1680.0	1687.5	13.6
T_3	Seed treatment with Saaf @ 2 g per kg seed	0.9(5.52)	2.0(8.13)	1.5(7.04)	88.9	1690.0	1661.0	1675.5	12.8
T_4	Seed treatment with Mencozeb @ 2 g per kg seed	6.3(14.50)	5.6(13.69)	6.0(12.92)	59.7	1650.0	1580.0	1615.0	8.2
T_5	Seed treatment with Carbendazim @ 2 g per kg seed	0.7(4.89)	1.7(7.49)	1.2(6.29)	91.9	1745.0	1690.0	1717.5	15.7
T_6	Seed treatment with Raxil @ 2 g per kg seed	1.9(7.92)	2.8(9.63)	2.3(8.72)	84.6	1680.0	1650.0	1665.0	12.1
T_7	Seed treatment with Tricyclazole @ 2 g per kg seed	6.2(14.46)	5.5(13.56)	5.9(14.06)	60.4	1625.0	1575.0	1600.0	7.7
T_8	Untreated control	11.2(19.54)	18.7(25.62)	14.9(22.71)		1510.0	1460.0	1485.0	
	Mean	3.7(11.09)	4.9(12.79)	4.3(11.79)		1668.1	1625.8	1646.9	

* Figures in parentheses are arc sin transformed values

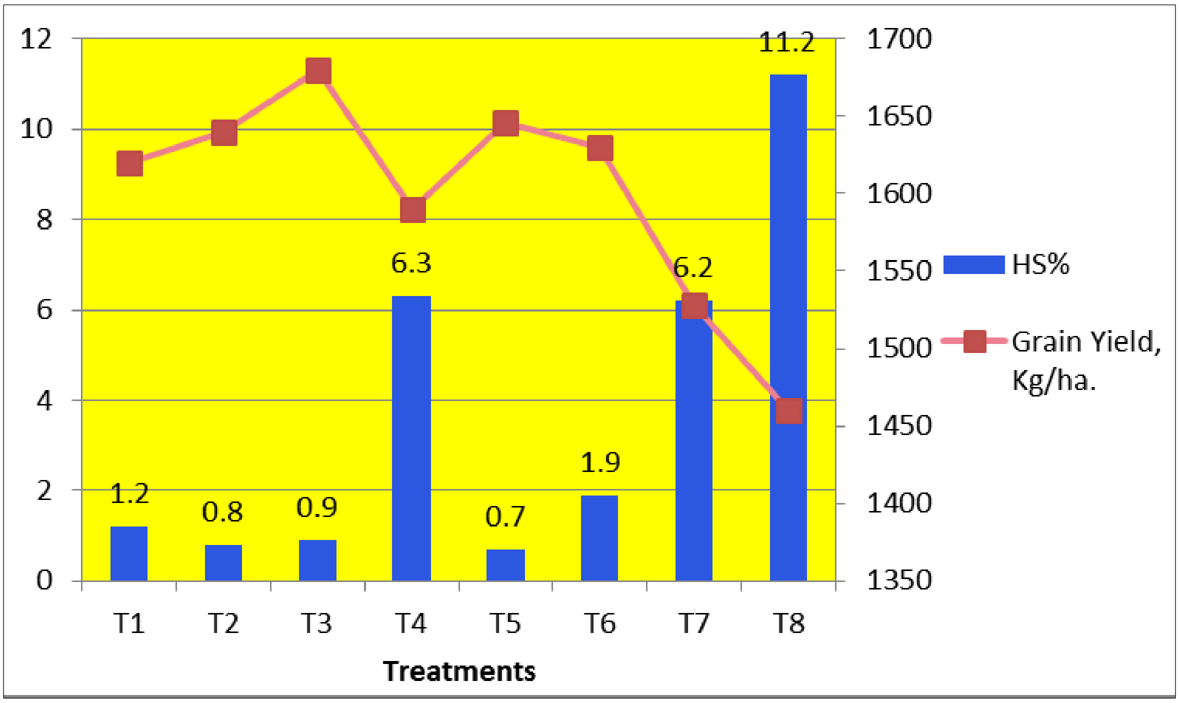

Fig 1: Head smut incidence (%) and grain yield (kg/ha.) in different treatments of Kodo millet (2010-11)

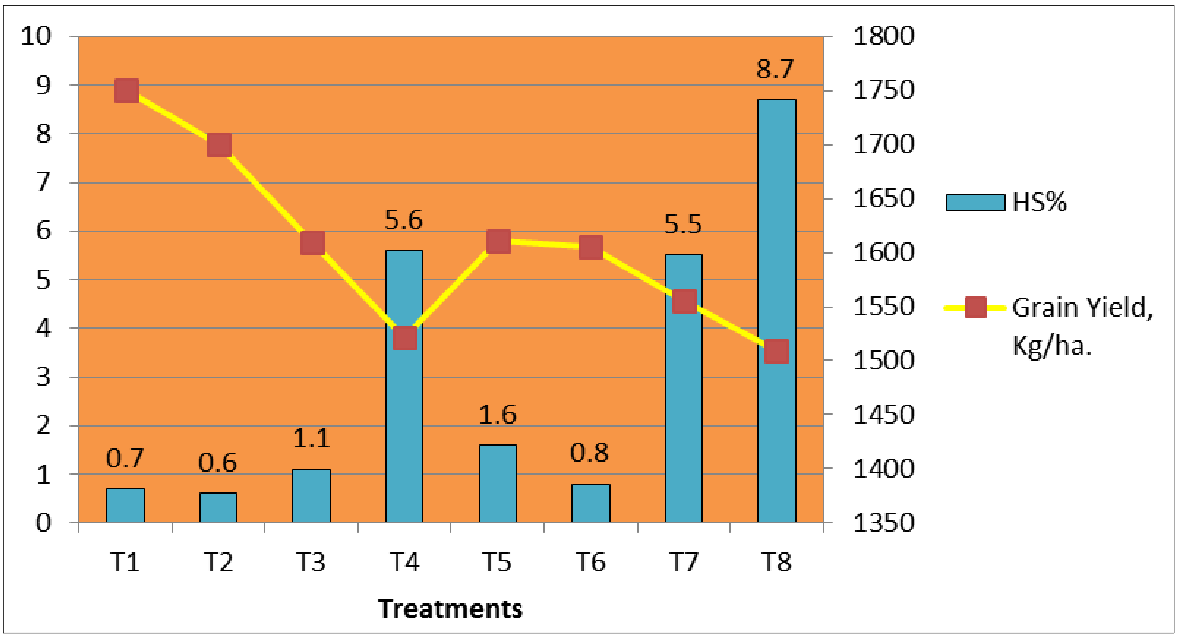

Fig 2: Head smut incidence (%) and grain yield (kg/ha.) in different treatments of Kodo millet (2011-12)

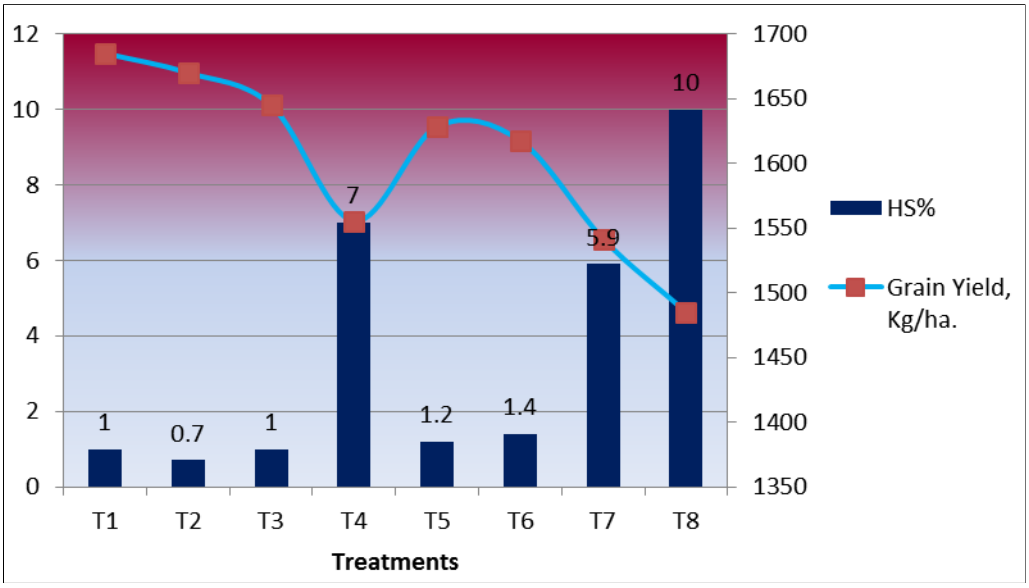

Fig 3: Average head smut incidence (%) and grain yield (kg/ha.) in different treatments of kodo millet (2010-11 and 2011-12)

Table 4: Teliospore germination of *Sorosporium paspali thunbergii* as influenced by fungicides at different concentrations

Treatments	Teliospore germination (%)				Mean	Percent inhibition% over control
	3 days	4 days	5 days	6 days		
Vitavax						
100 ppm	7.5(15.90)*	10.5(18.86)	12.8(20.91)	18.7(25.64)	12.4(20.62)	61.6
300 ppm	5.6(13.59)	8.3(16.76)	11.8(20.04)	14.5(22.35)	10.1(18.53)	68.7
500 ppm	0.0(0.28)	0.0(0.28)	4.7(12.52)	6.6(14.89)	2.8(9.63)	91.3
Mencozeb						
100 ppm	11.8(19.57)	14.2(22.10)	16.5(23.96)	22.4(28.24)	16.2(23.73)	49.8
300 ppm	8.9(17.30)	10.6(18.95)	13.4(21.42)	18.8(25.61)	12.9(21.05)	60.1
500 ppm	3.3(10.31)	8.3(16.73)	10.3(18.71)	11.3(19.64)	8.3(16.74)	74.3
Carbendazim						
100 ppm	9.8(18.22)	12.5(20.58)	14.1(22.00)	20.3(26.52)	14.2(22.14)	56.0
300 ppm	7.5(15.90)	9.3(16.65)	12.2(20.40)	16.8(24.22)	11.5(19.82)	64.4
500 ppm	0.0(0.28)	0.0(0.28)	5.6(13.69)	7.0(15.34)	3.1(10.14)	90.4
Control	20.5(26.89)	26.8(31.11)	35.6(36.64)	46.5(40.12)	32.3 (34.63)	
Mean	7.5(15.89)	10.0(18.44)	13.7(21.72)	18.3(25.33)	12.4(20.62)	
S.Em±	0.83	0.99	0.93	1.41		
CD=(0.05)	2.46	2.93	2.74	4.16		
CV	10.41	10.55	7.67	9.85		

* figures in parentheses are arc sin transformed values

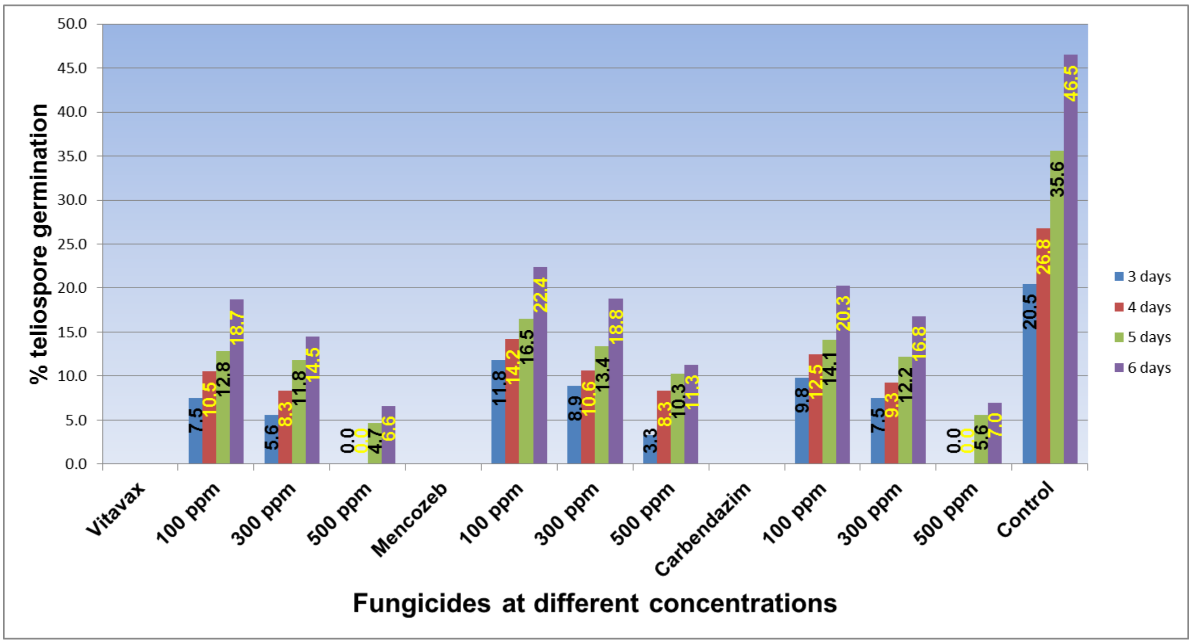

Fig 4: Percent teliospore germination of *Sorosporium paspali thunbergii* in various fungicides at 100, 300 and 500 ppm

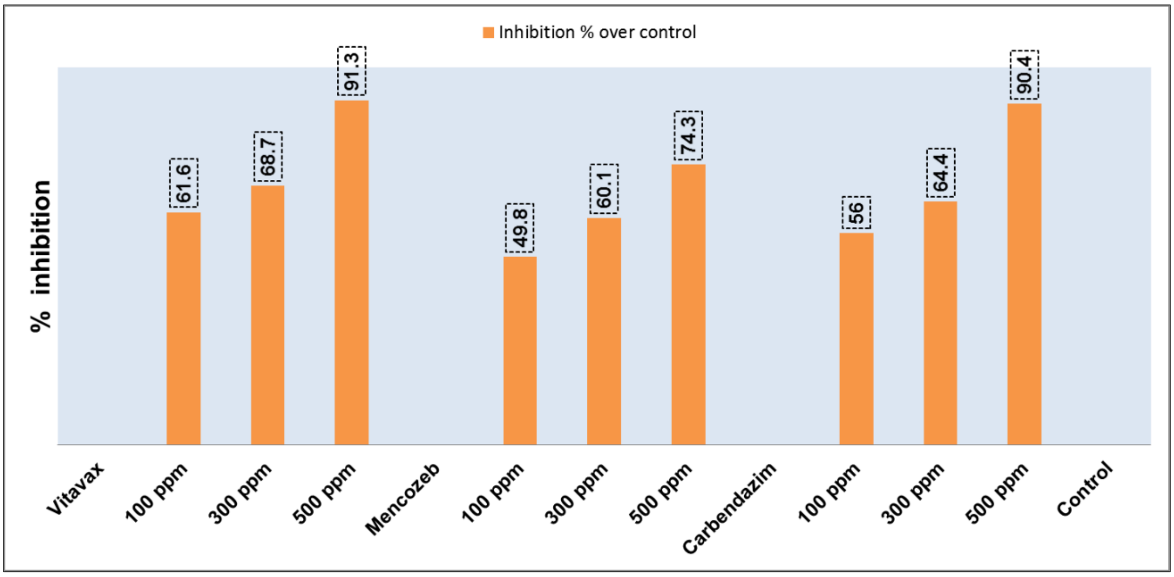

Fig 5: Average percent inhibition in teliospore germination of *Sorosporium paspali thunbergii* at different fungicidal concentration

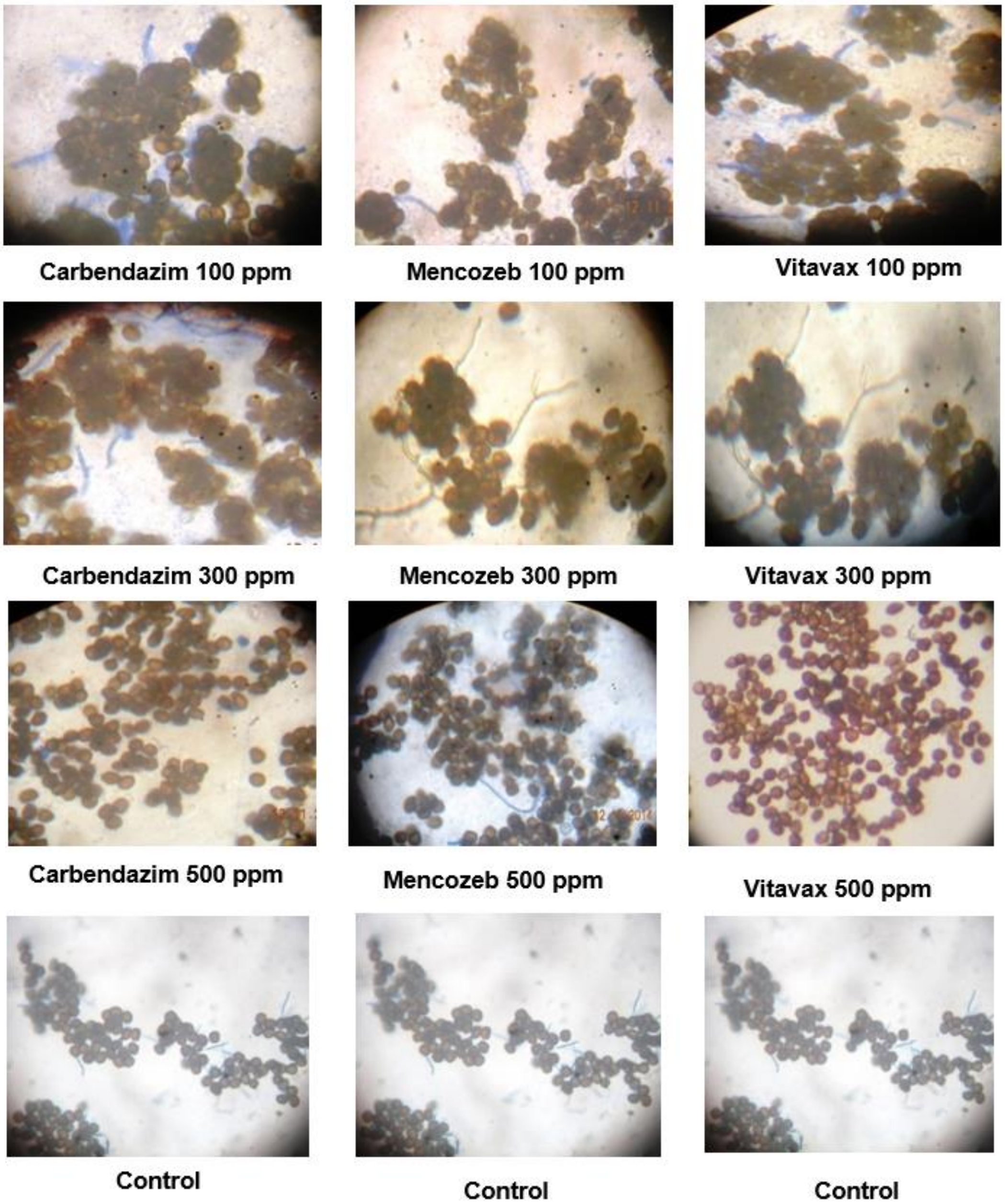

Plate 1: Teliospore germination of *Sosossporium paspali thunbergii a*t different fungicidal concentration

Conclusions

1. *In vivo*, carboxin@ 2 g kg^{-1} seed was found superior fungicide over rest of the fungicide which observed minimum head smut incidence (1.0%) and recorded maximum grain yield (1730.0 kg/ha.).
2. *In vitro*, carboxin was found most effective fungicide in reducing the teliospore germination at all the concentration 100, 300 and 500 ppm followed by carbendazim and mencozeb.

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